

CLAIMS

1. A transformed bryophyte cell that comprises i) a dysfunctional fucosyl transferase nucleotide sequence and ii) a
5 dysfunctional xylosyl transferase nucleotide sequence.
2. A transformed bryophyte cell according to claim 1 further comprising a nucleotide sequence operably linked to an exogenous promoter that drives expression in the said bryophyte cell
10 wherein said nucleotide sequence encodes a functional mammalian galactosyl transferase that is expressed in the bryophyte cell.
3. A transformed bryophyte cell according to claim 1 or claim 2 wherein the mammalian galactosyl transferase that is expressed
15 is a beta-1,4 galT.
4. A transformed bryophyte cell according to any one of claims 1 to 3 wherein the mammalian galactosyl transferase that is expressed is a human beta-1,4 galT.
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5. A transformed bryophyte cell according to any one of claims 1 to 4 wherein the said cell further comprises a nucleotide sequence operably linked to an exogenous promoter that drives expression in the said bryophyte cell wherein said nucleotide
25 sequence encodes a mammalian glycosylated polypeptide that is expressed in the bryophyte cell.
6. A bryophyte cell according to claim 5 wherein the bryophyte cell is selected from species of the genera *Physcomitrella*,
30 *Funaria*, *Sphagnum*, *Ceratodon*, *Marchantia* and *Sphaerocarpos*.
7. A bryophyte cell according to claim 6 wherein the bryophyte cell is selected from *Physcomitrella*.

8. A bryophyte cell according to claim 7 wherein the bryophyte cell is from *Physcomitrella patens*.

9. A bryophyte cell according to claim 8 wherein the mammalian glycosylated polypeptide is selected from the group comprising a polypeptide having a primary amino acid sequence of a human glycosylated polypeptide, a primary amino acid sequence of a non-human mammalian glycosylated protein, a primary amino acid sequence of an antibody or an active fragment thereof, and/or a primary amino acid sequence of a non-mammalian glycosylated polypeptide.

10. A bryophyte cell according to any one of claims 1 to 9 wherein the mammalian glycosylated polypeptide is a human polypeptide.

11. A bryophyte cell according to claim 9 or claim 10 wherein the mammalian glycosylated polypeptide is selected from the group consisting of human insulin, preproinsulin, VEGF, proinsulin, glucagon, interferons such as alpha-interferon, beta-interferon, gamma-interferon, blood-clotting factors selected from Factor VII, VIII, IX, X, XI, and XII, fertility hormones including luteinising hormone, follicle stimulating hormone growth factors including epidermal growth factor, platelet-derived growth factor, granulocyte colony stimulating, prolactin, oxytocin, thyroid stimulating hormone, adrenocorticotrophic hormone, calcitonin, parathyroid hormone, somatostatin, erythropoietin (EPO), and enzymes such as beta-glucocerebrosidase, haemoglobin, serum albumin, and collagen.

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12. A method of producing at least a bryophyte cell wherein fucT and xylT activity is substantially reduced that comprises introducing into the said cell i) a first nucleic acid sequence that is specifically targeted to an endogenous fucosyl transferase nucleotide sequence and ii) introducing into the

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said cell a second nucleic acid sequence that is specifically targeted to an endogenous xylosyl transferase nucleotide sequence.

- 5 13. A method according to claim 12 further comprising introducing into the said cell an isolated nucleic acid sequence that comprises nucleic acid operably linked to an exogenous promoter that drives expression in a bryophyte cell wherein said nucleic acid encodes a functional mammalian galactosyl
10 transferase polypeptide.
14. A method according to claim 12 or claim 13 wherein the galactosyl transferase nucleotide sequence is a beta-1,4 galactosyl transferase (beta-1,4 galT)nucleotide sequence.
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15. A method according to claim 14 wherein the galactosyl transferase nucleotide sequence is a human beta-1,4 galactosyl transferase (beta-1,4 galT)nucleotide sequence.
- 20 16. A method according to any one of claims 12 to 15 wherein the said transformed bryophyte cell further comprises a nucleotide sequence operably linked to an exogenous promoter that drives expression in the said bryophyte cell wherein said nucleotide sequence encodes a mammalian glycosylated polypeptide that is
25 expressed in the bryophyte cell.
17. A method according to any one of claims 12 to 16 wherein the mammalian glycosylated polypeptide is selected from the group comprising a protein having a primary amino acid sequence of a
30 human protein, a primary amino acid sequence of a non-human mammalian protein, a primary amino acid sequence of an antibody or an active fragment thereof, and/or a primary amino acid sequence of a non-mammalian protein.
- 35 18. A method according to any one of claims 12 to 17 wherein the glycosylated polypeptide is selected from the group consisting

of human insulin, preproinsulin, proinsulin, glucagon, interferons such as alpha-interferon, beta-interferon, gamma-interferon, blood-clotting factors selected from Factor VII, VIII, IX, X, XI, and XII, fertility hormones including
5 luteinising hormone, follicle stimulating hormone growth factors including epidermal growth factor, platelet-derived growth factor, granulocyte colony stimulating, prolactin, oxytocin, thyroid stimulating hormone, adrenocorticotrophic hormone, calcitonin, parathyroid hormone, somatostatin, erythropoietin
10 (EPO), and enzymes such as beta-glucocerebrosidase, haemoglobin, serum albumin, collagen, and human and non-human proteins selected from amidases, amylases, carbohydrases, cellulase, dextranase, esterases, glucanases, glucoamylase, lactase, lipases, pepsin, peptidases, phytases, proteases, pectinases,
15 casein, whey proteins, soya proteins, gluten and egg albumin.

19. A method according to any one of claims 12 to 18 wherein the bryophyte cell is selected from species of the genera
Physcomitrella, *Funaria*, *Sphagnum*, *Ceratodon*, *Marchantia* and
20 *Sphaerocarpos*.

20. A method according to claim 19 wherein the bryophyte cell is selected from *Physcomitrella*.

25 21. A method according to claim 20 wherein the bryophyte cell is from *Physcomitrella patens*.

22. A method according to any one of claims 12 to 21 wherein the mammalian glycosylated polypeptide is selected from the group
30 comprising a polypeptide having a primary amino acid sequence of a human glycosylated polypeptide, a primary amino acid sequence of a non-human mammalian glycosylated protein, a primary amino acid sequence of an antibody or an active fragment thereof, and/or a primary amino acid sequence of a non-mammalian
35 glycosylated polypeptide.

23. A method according to any one of claims 19 to 22 wherein the mammalian glycosylated polypeptide is a human polypeptide.

24. A method according to any one of claims 19 to 23 wherein the mammalian glycosylated polypeptide is selected from the group consisting of human insulin, preproinsulin, VEGF, proinsulin, glucagon, interferons such as alpha-interferon, beta-interferon, gamma-interferon, blood-clotting factors selected from Factor VII, VIII, IX, X, XI, and XII, fertility hormones including luteinising hormone, follicle stimulating hormone growth factors including epidermal growth factor, platelet-derived growth factor, granulocyte colony stimulating, prolactin, oxytocin, thyroid stimulating hormone, adrenocorticotrophic hormone, calcitonin, parathyroid hormone, somatostatin, erythropoietin (EPO), and enzymes such as beta-glucocerebrosidase, haemoglobin, serum albumin, and collagen.

25. A method according to any one of claims 12 to 24 wherein the exogenous promoter is selected from inducible, chemical-regulated, constitutive or cell specific promoters.

26. An isolated polynucleotide that encodes a functional mammalian glycosyl transferase for use in a method according to any one of claims 12 to 25.

27. An isolated polynucleotide according to claim 26 that encodes a recombinant mammalian galactosyl transferase for use in a method according to any one of claims 12 to 26.

28. An isolated polynucleotide according to claim 26 that encodes a recombinant human beta-1,4 galactosyl transferase for use in a method according to any one of claims 12 to 27.

29. A nucleic acid vector suitable for transformation of a bryophyte cell and including a polynucleotide according to any

one of claims 26 to 28.

30. A host cell containing a heterologous polynucleotide or nucleic acid vector according to any one of claims 26 to 29.

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31. A host cell according to claim 30 which is a bryophyte cell.

32. A host cell according to claim 29 which is a prokaryote cell.

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33. A method of producing a host cell according to any of claims 30 to 32, the method including incorporating said polynucleotide or nucleic acid vector into the cell by means of transformation.

15 34. Use of a polynucleotide according to any one of claims 26 to 28 in the production of a transgenic bryophyte cell.

20 35. A host cell according to claim 30 or 31 which is comprised in a bryophyte, or a bryophyte part, or an extract or derivative of a bryophyte or in a bryophyte cell culture.

36. A bryophyte plant or bryophyte tissue comprising a bryophyte cell according to any one of claims 30, 31, or 35.

25 37. A method of producing a bryophyte plant, the method including incorporating a polynucleotide or nucleic acid vector according to any of claims 26 to 29 into a bryophyte cell and regenerating a bryophyte from said cell.